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Root Necrosis Assessment of Plant Parasitic Nematodes of Banana (*Musa* spp.) at Arbaminch, Ethiopia

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Abstract
Of the extraordinary tourist destination place in our planet, Arba Minch, is a major producer of banana which is the most important food and cash crop in Ethiopia as well as in the world. Plant-parasitic nematodes are a primary pest of banana crop worldwide and are therefore a threat to food security. The study was conducted in Arba Minch, Ethiopia, to assess the level of root damage caused by plant parasitic nematodes of banana. During 2015, Twenty four composite banana root samples were collected from six randomly selected areas and transported to laboratory in Arba Minch University. And then, five functional root pieces per sample were assessed for necrosis, washed, surface dried, chopped, macerated and extracted using the modified Baermann funnel technique. Four genera of plant parasitic nematodes such as *Pratylenchus, Meloidogyne, Radopholus,* and *Rotylenchulus* were identified for the first time in Ethiopia. Necrosis observed in roots was moderate to high, for example, the lowest and highest percentage score on Giant and Dwarf Cavendish accounted 19.5% and 45% respectively. The maximum root necrosis recovered on Dwarf Cavendish (62%) indicates that farmers are suffering substantial losses due to several plant parasitic nematodes. Thus, development of nematode management tactics requires consideration of the damage caused by total phytonematode populations.
Keywords: Arba Minch, Banana, Cavendish, Ethiopia, Plant Parasitic Nematodes

1. Introduction

Banana (*Musa* spp.), originated in Malaysia (Heslop-Harrison and Schwarzacher, 2007), is one of the most important food and cash crop in Ethiopia as well as in the world. It is grown in tropical regions, especially in lowland areas where rainfall is in excess of 1250 mm per year (Gowen *et al*., 2005). Banana is the second major fruit crop next to citrus in Ethiopia. Among the potential banana producing areas, it is estimated that banana from Arba Minch, known for its good taste, has more than 80% of the market share in Ethiopia. However, yield per unit area of land is declining due to pests and diseases. Nematodes are among the primary pest of banana crop worldwide (Gowen and Quénéhérvé, 1990; Kashaija *et al*., 1994) and are therefore a threat to food security. The major plant parasitic nematodes such as *Radopholus similis*, *Meloidogyne javanica, Meloidogyne incognita, Helicotylenchus multicinctus, Pratylenchus coffeae, Pratylenchus goodeyi, Helicotylenchus dihystera and Rotylenchulus reniformis* have been reported from banana in Zimbabwe (Chitamba *et al*., 2013), Brazil (Lima *et al*., 2013), Congo (Kamira *et al.,* 2013) and Greece (Tzortzakakis, 2008). *Pratylenchus coffeae, Meloidogyne* species and *Helicotylenchus multicinctus* (Chau *et al*., 1997; Van Den Bergh, 2002) are the most important nematodes found in Vietnam on *Musa.* In a survey carried out in Tanzania by Sikora *et al*. (1989), 85% of the plants sampled had *P. goodeyi*, 60% contained *H. multicinctus* and 0-27% had *R. similis.* They are distributed across Africa where banana are grown, attack all banana cultivars and cause severe damage (Sarah, 2000). *Radopholus similis* and *Helicotylenchus multicinctus* are reportedly responsible for yield losses of 30-50% in Costa Rica, 40% in Africa, and exceeding 50% in East Africa (Speijer and Kajumba, 1996; Kashaija *et al*., 2004). Root-knot nematodes (*Meloidogyne* species) and the burrowing nematode (*R. similis*) can significantly weaken root systems, reduce yields, topple plant before harvest, make plant more prone to wind knockdowns, reduce fertilizer uptake and utilization, and reduce the banana-growing lifespan of a given piece of land (Nelson *et al*., 2006). These banana nematodes attack root and corm tissues causing damage that reduce bunch size, shorten the life of production, prolong the vegetative cycle and cause banana plants to topple (Chabrier and Quénéhérvé, 2003).

Earlier nematodes surveys conducted in Ethiopia have documented the presence of plant-parasitic nematode genera associated with Coffee such as *Helicotylenchus, Heterodera, Xiphinema, Rotylenchulus, Scutellonema, Tylenchorhynchus,* and *Criconemella* (Mekete *et al*., 2008), and with Enset such as *Pratylenchus* and *Aphelenchoides* (Bogale *et al*., 2004). However, neither plant-parasitic nematodes associated with banana nor damage caused by them was yet recorded in Ethiopia. The damage potential of these nematodes at farm level was not clear and documented and therefore needed to be investigated. Consequently, baseline information concerning the level of damage of plant parasitic nematodes of banana in Ethiopia was highly needed. It was crucial to design a research on assessment of damage levels for an effective banana nematode management in any given banana plantation. Thus, the objective of this study was to assess the level of root damage caused by plant parasitic nematodes of banana in Arba Minch, Ethiopia.

2. Materials and Methods

Study Area: The study was conducted from March to June 2015 at Arba Minch Zuria woreda, a major banana production area of Ethiopia. The centre of the study area, Arba Minch town, is located 505 kms South of Addis Ababa, the capital city of Ethiopia. It is located geographically at 37° 5`E longitude and 6°04` N latitude with altitude ranging 1200-1300 meters above sea level. The average annual rainfall ranges from 750-930 mm with mean average temperature of 30°c. The town is situated in well known East African Rift valley and surrounding by Lake Chamo and Abaya as well as the Nech Sar National Park (GZARDO, 2007). The main banana varieties grown in the area are Giant and Dwarf Cavendish.

Root Sampling: Depending on the farm size and types of varieties, 24 composite banana root samples were collected using a zig-zag pattern from six randomly selected banana fields: Amu farm, Chano, Limat, Kulfo, Sele and Shara. Each cultivar was sampled separately. The samples were collected following a procedure by Speijer and De Waele (1997), a hole of 20 x 20 x 20 cm was made adjacent roots and placed in a labelled plastic bag. Samples were taken from 5 to 8 randomly selected individual plants per field, pooled to form a composite root and transported to the laboratory within 24 hours of collection.

Root Necrosis Assessment: Root cortical necrosis (percentage) in banana roots was evaluated on five functional roots randomly selected per sample. The selected roots were cut to about 10cm length, each root piece sliced lengthwise and necrosis in the exposed cortical tissue scored as percentage, each root piece contributing a maximum score of 20% and for the five root pieces selected from the sample a total score of 100% (Speijer and De Waele, 1997).

Nematode Identification: The root pieces assessed for necrosis were washed with tap water, surface dried, chopped into 1 to 3 cm segments and thoroughly mixed before removing a 10 g fresh weight sample. The 10 g sub-sample was covered with water to just submerge the roots and the sample was macerated in a kitchen blender for 10-15 seconds. Nematodes from roots were extracted for 48hours using the modified Baermann funnel technique (Coyne *et al*., 2007). The extracted nematode suspensions were decanted into a beaker, allowed to settle for 2 hours and reduced to 25 ml by gently decanting off the excess water. Plant-parasitic nematodes were identified to genus and/or species level based on morphological characteristics using a compound microscope.

Data Analysis: Necrosis damage observations, expressed as percentages of root necrosis, were converted to root necrosis index (RNI%) using arcsin (x/100) for statistical analysis.

3. Results and Discussion

The most common genera of plant parasitic nematodes associated with banana were *Pratylenchus, Meloidogyne, Helicotylenchus, Radopholus,* and *Rotylenchulus* (Table 1). This agrees well with Chitamba *et al*. (2013) who reported that nematode species with different feeding habits usually exist as mixed populations in banana fields. Other workers (Gowen *et al*., 2005) also reported that nematode parasitism in the banana plantation are characterized by several nematode species causing simultaneous infections. These nematode genera detected are well known pathogens in banana roots (McSorley and Parrado, 1986; Sarah, 1989; Gowen, 1995; De waele and Davide, 1998; Marin *et al*., 1998).

Two banana varieties, named Dwarf Cavendish and Giant Cavendish, showed great variation in percentage of root necrosis ranged from 10% to 62%. Among these varieties, Dwarf Cavendish variety showed the highest average percentage of root necrosis accounted 45% while the lowest average percentage of root necrosis was recorded from Giant Cavendish that was 19.5%. Necrosis observed in roots was moderate to high, for example, the lowest and highest percentage score on banana was 10% and 62% at Sele and Kulfo, respectively (Table 1).

Similarly, Moens *et al*. (2001) found a significant correlation between *R. similis* and root necrosis or damage, ranging from 0.62 to 0.75, in root samples of banana plantations. This was also coincided with those described by Pinochet (1978) who said that *Pratylenchus* species damaged banana roots and reduced yield. *Helicotylenchus multicinctus* (McSorley and Parrado, 1986; Davide, 1996; Mani and Al Hinai, 1996; Chau *et al*. 1997) damaged the banana root system and reduce yield by 19% (Speijer and Fogain, 1999) to 34% (Reddy, 1994). In Florida, *H. multicinctus* can cause severe root damage resulting in the toppling mature plants (McSorley, 1986; Gowen, 1995). In Lebanon, *H. multicinctus* is considered the most important banana root parasitic nematode (Sikora and Schlösser, 1973). The same authors felt that this nematode, together with *M. incognita*, was associated with the general decay of the banana root system observed. Not only damaging the root of banana but also nematodes reduce bunch weight and plant longevity, and increase the crop cycle duration (Quénéhérvé, 1991). In Malaysia, Razak (1994) reported stunted growth and small bunches due to the infection

of *M. incoginta*. Similar effects on plant growth have been reported in India (Patel *et al*., 1996) where a delay in flowering was also observed. Van den Bergh *et al*. (2000) reported that *Meloidogyne* species had the most negative effect on the bunch characteristics of banana plants in Vietnam.

In this finding, percentage of root necrosis due to nematodes differed among banana genotypes and locations. These results agree with Speijer *et al*. (1994) who observed that overall nematode damage levels displayed high variability among different sites in Uganda. In contrast, Kamira *et al.* (2013) reported that damage to banana roots (percentage necrotic root tissue) was not significantly different among banana types. The percentage of root necrosis was greater than 5% in the study areas even though Speijer *et al*. (1994) considered nematodes damage to be low when the necrosis of the root cortex did not exceed 5% on primary roots of banana. The highest percentage of root damage was due to nematodes which spread as the result of irrigation water, lack of management practices, contaminated suckers for planting materials and harvested plant roots remained in soil for many years in the study area. Similarly, many authors considered that nematodes are mainly disseminated by water (Bur and Robinson, 2004).

Table 1. Nematode damage (root necrosis) and distribution of plant parasitic nematodes associated with two banana varieties in Arba Minch Zuria Woreda

Nematode Eco-region	Cavendish cultivars	RNI%	*Rotylenchus reniforms*	*Radopholus similis*	*Pratylenchus coffeae*	*Helicotylenchus multicinctus*	*Meloidogyne incognita*
Kulfo (3)	Dwarf	0.62	+	+	+	+	+
Limat (3)	Dwarf	0.6	+	+	+	+	+
Chano (4)	Dwarf	0.53	-	+	-	+	+
Shara (3)	Dwarf	0.4	+	+	+	+	+
Limat (2)	Giant	0.25	-	+	+	+	+
Amu Farm (3)	Giant	0.22	+	+	-	+	+
Chano (2)	Giant	0.18	-	+	+	+	+
Sele (3)	Giant	0.13	-	+	-	+	+
Sele (1)	Dwarf	0.1	-	+	-	-	-

Note: Figures in the parenthesis are the total number of samples collected; (+) Denotes occurrence of nematodes, (-) Denotes did not yield any nematodes. Data of percentage of root necrosis were converted to arcsin (x/100) for statistical analysis. RNI = root necrosis index.

The destruction of the root system probably explains why plants infested with nematodes normally topple (Barkeye *et al*., 2000). This implies that banana root damage by plant parasitic nematodes could be a very useful tool to identify resistance or tolerance in varietal screening trials. Similarly, a variety with severe damage score which records significant yields could be a tolerant genotype. Based on the extent of the necrosis, considerable losses also can be expected (Peregrine and Bridge, 1992). Damage score usually has a strong relationship with crop yield losses (Coyne *et al.,* 2007). Severely damaged roots normally topple-over at the expense of yield, while undamaged root systems have the capacity to support fruit bearing plants till harvest.

The highest root necrosis recovered on Dwarf Cavendish variety (62%) indicates that farmers are suffering substantial losses due to nematodes, which are undoubtedly being spread as the result of lack of management practices and contaminated suckers for planting materials, which are a practice common in may smallholder banana growing areas (Tenkouana *et al*., 2006; Kamira *et al*., 2013). This also showed the relatively high susceptibility and sensitivity to plan parasitic nematodes of banana genotypes grown by the farmers in the study areas. In addition, most growers are unaware that nematodes are a cause of banana production problems (Brooks, 2004).

3. Conclusion

In conclusion, this work showed that a mixture of plant parasitic nematodes damaged bananas roots in Arba Minch, Ethiopia. Therefore, development of nematode management tactics requires consideration of the damage caused by total phytonematode populations. These finding also identified nematode infected suckers which were ready to be transported to other part of the country. Thus, selecting nematode free planting material (suckers) planted on fields not previously grown under bananas would be a good starting point.

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